Influence of level of supplemental whole flaxseed on forage intake and site and extent of digestion in beef heifers consuming native grass hay

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Influence of level of supplemental whole flaxseed on forage intake and site and extent of digestion in beef heifers consuming native grass hay[1,2]

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ABSTRACT: The objectives of this study were to evaluate the influence of supplemental whole flaxseed level on intake and site and extent of digestion in beef cattle consuming native grass hay. Nine Angus heifers (303 ± 6.7 kg of BW) fitted with ruminal and duodenal cannulas were used in a triplicated 3 × 3 Latin square. Cattle were given ad libitum access to chopped native grass hay (9.6% CP and 77.5% NDF, OM basis). All animals were randomly allotted to 1 of 3 experimental treatments of hay plus no supplement (control); 0.91 kg/d whole flaxseed (23.0% CP, 36.3% NDF, and 25.5% total fatty acid, OM basis); or 1.82 kg/d whole flaxseed on a DM basis. Supplemental flaxseed tended to decrease (linear, $P = 0.06$) forage OM intake. However, total OM intake did not differ ($P = 0.29$) with increasing levels of flaxseed. Total duodenal OM flow increased (linear, $P = 0.05$) with additional flaxseed in the diet, and no differences ($P = 0.29$) were observed for microbial OM flow. True ruminal OM disappearance was not affected ($P = 0.14$) by supplemental flaxseed. Apparent lower tract OM digestibility increased (linear, $P = 0.01$) with level of whole flaxseed. Apparent total tract OM digestibility was not different ($P = 0.41$) among treatments. Nitrogen intake increased (linear, $P < 0.001$) with supplemental flaxseed. In addition, total duodenal N flow tended ($P = 0.08$) to increase with additional dietary flaxseed. However, true ruminal N digestibility did not differ ($P = 0.11$) across treatment. Supplemental whole flaxseed did not influence ruminal ($P = 0.13$) or total tract ($P = 0.23$) NDF digestibility. Ruminal molar proportion of propionate responded quadratically ($P < 0.001$) with increasing levels of whole flaxseed. An increase in the duodenal supply of 18:3n-3 ($P < 0.001$), total unsaturated fatty acids ($P < 0.001$), and total fatty acids ($P < 0.001$) was observed with additional dietary whole flaxseed. Apparent postruminal 18:3n-3 disappearance tended to decrease ($P = 0.07$) as intake of flaxseed increased. Overall, the inclusion of 1.82 kg/d of flaxseed does not appear to negatively influence nutrient digestibility of a forage-based diet and therefore can be used as an effective supplement to increase intestinal supply of key fatty acids important to human health.

Key words: beef cattle, digestion, fatty acid, flaxseed, forage

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INTRODUCTION

Feeding beef cattle diets high in n-3 fatty acids is warranted for livestock producers interested in enhancing the human healthfulness of meat products (Weill et al., 2002) or reducing reproductive losses (Ambrose et al., 2006). Feeding flaxseed, which is high in linolenic acid (56% 18:3n-3), is a viable option for bolstering the n-3 fatty acid concentration in livestock diets and subsequent n-3 fatty acid concentration in ruminant products (Soita et al., 2003; Kronberg et al., 2006; Maddock et al., 2006). Unfortunately, there are limitations on the level of fat that can be added to ruminant diets because of reductions in intake and digestibility (Schauff and Clark, 1992). This is especially true for fats high in PUFA (Palmquist and Jenkins, 1980; Jenkins, 1993). However, flaxseed seems to be unique in this regard because previous work by Zhang et al. (2007) showed an increase in DM digestibility for lactating ewes fed a silage-based diet containing 8% flaxseed compared with diets with 7.3% canola seeds or no oilseeds. In feedlot diets, flaxseed does not influence (Maddock et

[1]Mention of a proprietary product does not constitute a guarantee or warranty of the product by USDA or the authors and does not imply its approval to the exclusion of another product that may also be suitable. USDA, ARS, Northern Plains Area, is an equal opportunity/affirmative action employer. All agency services are available without discrimination.

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al., 2006) or increases (Drouillard et al., 2004) dietary intake, and an overall improvement in ADG is observed with inclusions of 5 to 8% flaxseed in the diet. However, no information exists with flaxseed fed in high-forage diets. Nevertheless, others have evaluated the use of oilseeds in forage-based diets and have found varying results for intake and total-tract digestibility of DM (Albro et al., 1993) and OM (Leupp et al., 2006). Specifically, Albro et al. (1993) fed weanling beef steers 1.5 kg/d of whole soybeans and did not observe a difference in intake but did report an increase in total-tract DM digestibility. Leupp et al. (2006) did not observe any differences in either forage intake or total-tract OM digestibility when canola (whole or ground) was offered to steers fed low-quality grass hay. Therefore, our objectives were to evaluate site and extent of digestion in beef heifers fed increasing amounts of whole flaxseed and consuming native grass hay.

MATERIALS AND METHODS

All experimental procedures were reviewed and approved by the Northern Great Plains Research Laboratory, Animal Care and Use Committee.

Animals and Diets

Nine Angus heifers (average BW: 303 ± 6.7 kg) fitted with ruminal and J-style duodenal cannulas (manufactured by the USDA-ARS, Northern Great Plains Research Laboratory) were used in a triplicated 3 × 3 Latin square. Animals were housed in a temperature-controlled barn within 3.3- × 2.7-m pens equipped with cup waterers. Before the initiation of the experiment, cattle were offered chopped (5 cm) native grass hay [Kentucky bluegrass (*Poa pratensis* L.), smooth brome (*Bromus inermus* Leyss.), blue grama (*Bouteloua gracilis* (H. B. K.) Griffiths), needle and thread (*Stipa comata* Trin. and Rupr.), green needlegrass (*Stipa viridula* Trin.), western wheatgrass, and sedges (*Carex* spp.); 9.6% CP and 77.5% NDF, OM basis; Table 1] at 5% above the previous day's intake and were given ad libitum access to whole flaxseed for 21 d in an effort to determine the level at which the animal would freely consume flaxseed. All pens were equipped with large feed bunks for hay, and bucket holders were mounted so that intake of hay and flaxseed could be measured individually. It was determined that the average maximal amount of whole flaxseed consumed was 1.82 kg/d. Therefore, cattle were fed hay at 5% above the previous day's intake and randomly allotted to 1 of 3 experimental treatments of hay plus no supplement; 0.91 kg/d of whole flaxseed on a DM basis; or 1.82 kg/d of whole flaxseed on a DM basis. Daily sampling of hay, flaxseed, and orts started on d 12 and continued through d 21 of each period. Heifers were fed twice daily at 0600 and 1800 h and given free access to trace mineralized salt (American Stockman Trace Mineralized Salt, North American Salt Co., Overland Park, KS: NaCl >95.5%; Zn >3,500 mg/kg; Fe >2,000 mg/kg; Mn >1,800 mg/kg; Cu >280 mg/kg; I >100 mg/kg; Co >60 mg/kg). Each experimental period was 21 d with a 17-d adaptation to ensure adequate adjustment of the gastrointestinal tract to the new dietary treatment and 4 d of intensive sampling. Starting on d 8 of each experimental period, gelatin boluses (size #11, Torpac Inc., Fairfield, NJ) containing 5 g of TiO_2 were placed into the central rumen twice daily at each feeding as an external marker for digesta flow (Myers et al., 2004).

Table 1. Chemical composition of chopped grass hay and whole flaxseed fed to beef heifers[1]

Item	Chopped grass hay	Whole flaxseed
DM	94.5	93.1
OM, % of DM	90.3	96.6
N, % of OM	1.53	3.82
NDF, % of OM	77.6	36.5
Fatty acid, % of OM		
16:0	0.335	1.48
18:0	0.041	0.89
18:1n-9	0.079	4.48
18:2n-9	0.188	4.55
18:3n-3	0.263	14.9
Total	0.961	26.5

[1]Samples for nutrient analysis were taken on d 12 through 21 of each period. Chemical composition values are reported as means of 3 periods.

Sampling

Beginning at 0400 h on d 18 of the experimental period, duodenal (200 mL) and fecal (50 mL) samples were collected every 4 h. On d 19 of the sampling period, duodenal and fecal collection times were advanced 2 h so that samples were collected to represent every 2 h in a 24-h period. Fecal samples were composited (equal volume) over time by heifer and dried in a 55°C forced-air oven, ground (Wiley mill, 1-mm screen; Arthur H. Thomas, Philadelphia, PA), and composited. Duodenal digesta samples were composited (equal volumes) within heifer for each period and immediately frozen. Duodenal digesta samples were lyophilized (Freezemobile 25SL Freeze Dryer, The VirTis Co., Gardiner, NY) and ground (Wiley mill; 1-mm screen).

Immediately before the 0600 h feeding on d 20, whole ruminal contents (500 mL) were removed and 200 mL of Co-EDTA (Uden et al., 1980) was dosed intraruminally (0 h). Whole ruminal contents were collected at 3, 6, 9, 12, 15, 18, 21, 24, and 36 h postdosing (samples collected at 24 and 36 h were analyzed for Co only). Immediately after collection, ruminal pH was measured using a combination electrode (Orion Research Inc., Boston, MA), contents were strained through 4 layers of cheesecloth, and a 10-mL aliquot of strained ruminal fluid was acidified with 0.1 mL of 3.6 *M* H_2SO_4 and immediately frozen at −20°C. In addition, an unstrained sample of whole ruminal contents was placed in a blender (Hamilton Beach/Proctor Silex, Washington,

NC) with an equal volume of 0.9% NaCl (wt/vol) solution and homogenized for 1 min to dislodge particulate-associated bacteria. The homogenate was then strained through 8 layers of cheesecloth and immediately frozen for subsequent bacterial isolation by differential centrifugation (Merchen et al., 1986).

Laboratory Analysis

All feed, orts, microbes, duodenal digesta, and fecal samples were analyzed for DM and ash (AOAC, 1990). Nitrogen content of feed, microbes, duodenal digesta, and feces were determined using a Carlo Erba Model NA 1500 Series 2 N/C/S analyzer (CE Elantech, Lakewood, NJ). Neutral detergent fiber of feed, duodenal digesta, and feces were determined using an Ankom 200 fiber analyzer (Ankom Technology, Fairport, NY). Duodenal and fecal samples were analyzed for TiO_2 according to the procedures of Myers et al. (2004) using a spectrophotometer (DU−640, Beckman Instruments Inc., Fullerton, CA). Microbial and duodenal samples were analyzed for purines as described by Zinn and Owens (1986).

Ruminal fluid samples were centrifuged at 20,000 × *g* for 20 min and a 2.5-mL aliquot was added to 0.5 mL of 25% (wt/vol) metaphosphoric acid containing 2 g/L of 2-ethyl-butyric acid (Goetsch and Galyean, 1983). These samples were analyzed for concentrations of VFA using a Varian 3800 gas chromatograph (Varian Inc., Palo Alto, CA) equipped with a 15 m × 0.533 mm (i.d.) column (Nukol, Supelco, Bellefonte, PA) with an initial column temperature of 80°C held for 1 min, ramped to 140°C at 20°C/min, and then ramped to 175°C at 30°C/min. Injector and detector temperatures were held at 250°C. The column flow rate was 8 mL/min and H_2 was the carrier gas. Approximately 100 mg of duodenal digesta was reconstituted to 3% DM using 0.1 *N* HCl for subsequent analysis of NH_3 concentration. Concentration of NH_3 in ruminal fluid and reconstituted duodenal digesta was determined using the phenol-hypochlorite procedure of Broderick and Kang (1980). Ruminal fluid Co concentrations were determined by atomic absorption spectroscopy using an air/acetylene flame (Model 3110, Perkin Elmer Inc., Norwalk, CT).

Feed was analyzed for fatty acids via direct transesterification (Whitney et al., 1999) with methanolic-HCl (Kucuk et al., 2001), and duodenal digesta fatty acids were analyzed for fatty acid using the procedures of Lake et al. (2006). Separation of fatty acid methyl esters was achieved by GLC (Model CP-3800, Varian Inc.) with a 100 m × 0.25 mm (i.d.) × 0.2 μm (film thickness) capillary column (SP-2560, Supelco) and H_2 gas as the carrier at 1.0 mL/min for feedstuffs and 1.5 mL/min for duodenal digesta. Initial oven temperature was maintained at 120°C for 2 min, ramped to 210°C at 6°C/min, and then ramped to 250°C at 5°C/min. Injector temperature was 260°C and flame-ionization detector temperature was 300°C. Identification of peaks was accomplished using purified fatty acid standards (Sigma-Aldrich, St. Louis, MO; Nu-Chek Prep, Elysian, MN; Matreya, Pleasant Gap, PA). Furthermore, based on the chromatogram published by Loor et al. (2004), a peak appearing between peaks confirmed to be 18:1 *trans*-11 and 18:1n-9 was putatively identified as 18:1 *trans*-13+14.

Calculations and Statistical Analysis

Nutrient flows, fluid passage rate, and microbial efficiency were calculated as described by Scholljegerdes et al. (2004b). Furthermore, biohydrogenation of 18C unsaturated fatty acids was calculated by using the equations of Tice et al. (1994) and Scholljegerdes et al. (2004a).

All data were analyzed using the MIXED procedure (SAS Inst. Inc., Cary, NC) as a triplicated 3 × 3 Latin square experiment. The model included animal as the random variable. In addition, time course data included the effects of animal, period, treatment, time, and treatment × time. Autoregression order one was determined to be the most desirable covariance structure according to the Akaike's information criterion. Orthogonal contrasts were used to compare linear and quadratic responses to level of flax intake (Steel and Torrie, 1980). There was a treatment × time interaction for ruminal NH_3 ($P < 0.001$) and the molar proportion of isovalerate ($P = 0.02$), whereas no other treatment × time interactions were observed ($P \geq 0.27$) for other time course data; therefore, only main effects are presented. A significance level of 0.05 was used to separate treatment effects.

RESULTS AND DISCUSSION

Intake and Digestibility of OM

The addition of whole flaxseed tended to reduce forage OM intake (linear, $P = 0.06$), but total OM intake did not differ ($P = 0.29$) because of the inclusion of whole flaxseed (Table 2). The substitution rate was a 0.65-kg decrease in forage intake per 1-kg increase in flaxseed consumed (forage intake = −0.647 flaxseed intake + 6.35, $R^2 = 0.13$; $P = 0.07$; data not shown). Leupp et al. (2006) did not report any differences in OM intake of low-quality grass hay or total intake with supplemental canola seeds. Pavan et al. (2007) reported a decrease in both forage and total DM consumption when level of corn oil increased for beef steers grazing endophyte-free tall fescue. Despite no differences being observed for total OM intake, duodenal OM flow increased ($P = 0.05$) linearly with supplemental flaxseed. However, no differences ($P = 0.29$) were observed for microbial OM flow to the duodenum. When ruminal digestibility is presented as a percentage of intake, no differences ($P = 0.14$) were observed for true ruminal OM digestibility across flaxseed intake levels. This is contrary to other reports (Doreau and Chilliard, 1997b; Scholljegerdes et al., 2004a) that reported a decrease in true

Table 2. Influence of supplemental whole flaxseed level (kg/d) on OM intake, flow, and digestibility in beef heifers consuming native grass hay

	Treatment[1]				Contrast[3]	
Item	0	0.91	1.82	SEM[2]	L	Q
OM intake, g/d						
Forage	6,358	5,771	5,236	435	0.06	0.96
Total	6,358	6,636	6,966	436	0.29	0.96
OM flow, g/d						
Duodenal	3,963	4,275	4,721	254	0.05	0.83
Microbial	1,127	1,346	1,208	138	0.67	0.29
Fecal	2,461	2,548	2,633	161	0.45	1.0
OM digestibility						
True ruminal, % of intake[4]	54.4	54.5	49.0	3.2	0.14	0.37
Lower tract, % of duodenal flow	37.6	40.9	44.1	1.6	0.01	0.97
Total tract, % of intake	60.1	61.7	62.1	1.7	0.41	0.75

[1]Treatments: 0 = ad libitum chopped native grass hay only; 0.91 = ad libitum chopped native grass hay plus 0.91 kg/d whole flaxseed (DM basis); 1.82 = ad libitum chopped native grass hay plus 1.82 kg/d of whole flaxseed (DM basis).
[2]n = 9.
[3]L = linear; Q = quadratic.
[4]Corrected for microbial OM.

ruminal OM digestibility, yet agrees with Leupp et al. (2006) who did not observe a depression in ruminal OM digestibility of forage-based diets supplemented with canola. It is not clear why these differences occurred between experiments. It is not likely due to differences in dietary fatty acid percentage, because Scholljegerdes et al. (2004a) fed diets ranging from 3.2 to 8.0% total fatty acids and Doreau and Chilliard (1997b) fed diets that ranged from 0.77 to 3.5% total fatty acids. Leupp et al. (2006) fed canola at 8% of DMI, which provided animals with diets that were approximately 4.2% total fatty acids. In the current experiment, feeding heifers 0.91 or 1.82 kg/d of whole flaxseed led to dietary contributions from the flaxseed of 13.0 and 24.8% of the daily OM intake. Therefore, dietary total fatty acid percentage ranged from 0.96 to 7.4. Because our highest level of fat inclusion was greater than either of the previously mentioned studies, it is difficult to conclude that fat intake level was the cause of discrepancy among these previous reports (Doreau and Chilliard, 1997b; Scholljegerdes et al., 2004a; Leupp et al., 2006) and the current experiment. It should be noted that Doreau and Chilliard (1997b) and Leupp et al. (2006) only reported crude fat; therefore, dietary total fatty acid percentage was calculated from crude fat using the equation of Allen (2000):

$$[\%\ \text{fatty acid} = -0.98 + (1.03 \times \%\ \text{ether extract})].$$

One could attribute these differences to availability of supplemental fat. In the current experiment, flaxseed was fed as the whole seed, whereas Doreau and Chilliard (1997b) fed fish oil and Scholljegerdes et al. (2004a) fed cracked safflower seeds, which would allow greater availability of fatty acids in the rumen. However, Leupp et al. (2006) fed both whole and ground canola and again found no differences in ruminal OM digestibility. Therefore, processing does not seem to be the reason for the inconsistencies across these experiments.

Apparent lower tract OM digestibility (% of duodenal flow) linearly increased ($P = 0.01$) when flax was added to the diet (Table 2). These findings agree with previous work in which feeding flaxseed to dairy cows consuming a 55:45 forage:concentrate ratio also increased lower tract OM digestibility (Gonthier et al., 2004). Despite the increase in lower tract digestibility in the current experiment, apparent total-tract OM digestibility (% of intake) did not differ ($P = 0.41$) across dietary treatments. This is contrary to previous results that reported an increase in total-tract digestibility in dairy cows fed linseed oil (Ueda et al., 2003) and flaxseed (Gonthier et al., 2004) in a total mixed ration.

Intake and Digestibility of N

Overall, N intake increased linearly ($P < 0.001$) with additional flaxseed (Table 3). However, supplemental flaxseed only tended (linear, $P = 0.08$) to increase the supply of N reaching the duodenum. Duodenal supply of microbial N and NH_3 did not differ ($P \geq 0.22$) across treatments. Others have indicated that duodenal microbial N supply (g/d) was unaffected by fat feeding (Brokaw et al., 2001; Scholljegerdes et al., 2004a) with forage-based diets. Nonammonia, nonmicrobial N flow responded quadratically ($P = 0.03$), with heifers fed 0.91 kg/d of whole flaxseed being lower than heifers fed 1.82 kg/d, which had the greatest duodenal N supply. This quadratic effect indicates that feeding 0.91 kg/d of whole flaxseed may increase ruminal degradability of dietary N. A quadratic tendency ($P = 0.10$) was observed for true ruminal N digestibility. Likewise, Murphy et al. (1987) were unable to detect a linear response in ruminal N digestibility when lactating dairy cows consuming a 59:41 forage:concentrate were fed 0, 1, or 2 kg of rapeseed and although not reported as a

Table 3. Influence of supplemental whole flaxseed level (kg/d) on N intake, flow, and digestibility (OM basis) in beef heifers consuming native grass hay

	Treatment[1]				Contrast[3]	
Item	0	0.91	1.82	SEM[2]	L	Q
N intake, g/d	97.9	121.2	146.7	6.8	<0.001	0.88
Duodenal N flow, g/d						
Total	136	146	162	10	0.08	0.85
Microbial	91	108	85	14	0.72	0.22
NH_3	10.7	13.4	11.9	1.3	0.53	0.23
NANM[4]	33.5	24.8	65.1	11	0.02	0.03
Fecal N flow, g/d	48.4	48.6	47.2	3.9	0.75	0.79
N digestibility						
True ruminal, % of intake[5]	66.4	76.9	55.1	9.8	0.30	0.10
Lower tract, % of duodenal flow	64.2	66.0	70.7	0.02	0.003	0.38
Total tract, % of intake	49.2	60.4	67.9	2.6	<0.001	0.50
MOEFF[6]	29.1	28.7	25.6	3.0	0.42	0.73

[1]Treatments: 0 = ad libitum chopped native grass hay only; 0.91 = ad libitum chopped native grass hay plus 0.91 kg/d whole flaxseed (DM basis); 1.82 = ad libitum chopped native grass hay plus 1.82 kg/d of whole flaxseed (DM basis).
[2]n = 9.
[3]L = linear; Q = quadratic.
[4]Nonammonia, nonmicrobial N.
[5]Corrected for microbial N and NH_3.
[6]MOEFF = g of microbial N/kg of OM truly fermented.

significant quadratic effect, cattle fed 1 kg of rapeseed had numerically greater ruminal N digestibility than those fed 2 kg of rapeseed. Apparent lower tract N digestibility increased ($P = 0.003$) with level of flaxseed. These data would indicate that whole flaxseed did not hinder ruminal N metabolism and that N escaping ruminal degradation is available at the small intestine. Apparent total-tract N digestibility was greater ($P < 0.001$) for heifers supplemented with whole flaxseed.

Intake and Digestibility of NDF

No differences were observed ($P \geq 0.20$) for NDF intake or duodenal and fecal NDF flow (Table 4). In turn, the inclusion of whole flaxseed had no effect on ruminal, lower tract, or total-tract NDF digestibility ($P \geq 0.13$). Inclusion of flaxseed in the current experiment was 3.2 and 5.8% of added dietary fatty acids (DM basis) for the 0.91 or 1.82 kg/d treatments, respectively. Therefore, the fat offered in this experiment is below the amount of added fat (6.3%) found to inhibit fiber digestibility (Moore et al., 1986). An alternative explanation would be that by feeding whole oilseeds we provided partial protection of the fatty acids, which can be an effective way to avoid the negative effects of fat on fiber digestion (Murphy et al., 1987). However, this is unlikely because of the relatively extensive biohydrogenation of 18C fatty acids observed in this trial.

Ruminal Fermentation Patterns

Ruminal pH tended (linear, $P = 0.06$) to increase linearly with increasing levels of whole flaxseed (Table 5). There was a treatment × time interaction ($P < 0.001$) for ruminal NH_3 (data not shown). This interaction occurred due to a reduction in the difference between

Table 4. Influence of supplemental whole flaxseed level (kg/d) on NDF intake, flow, and digestibility (OM basis) in beef heifers consuming native grass hay

	Treatment[1]				Contrast[3]	
Item	0	0.91	1.82	SEM[2]	L	Q
NDF intake, g/d	4,935	4,776	4,689	341	0.57	0.92
Duodenal NDF flow, g/d	2,379	2,120	2,349	147	0.89	0.20
Fecal NDF flow, g/d	1,863	1,724	1,672	119	0.26	0.76
NDF digestibility						
Ruminal, % of intake	49.8	55.0	49.1	3.0	0.87	0.13
Lower tract, % of duodenal flow	21.0	19.4	27.3	0.6	0.22	0.28
Total tract, % of intake	60.5	64.3	64.0	2.0	0.23	0.41

[1]Treatments: 0 = ad libitum chopped native grass hay only; 0.91 = ad libitum chopped native grass hay plus 0.91 kg/d whole flaxseed (DM basis); 1.82 = ad libitum chopped native grass hay plus 1.82 kg/d of whole flaxseed (DM basis).
[2]n = 9.
[3]L = linear; Q = quadratic.

Table 5. Influence of supplemental whole flaxseed level (kg/d) on ruminal pH, NH_3, fluid passage rate, and VFA in beef heifers consuming native grass hay

	Treatment[1]				Contrast[3]	
Item	0	0.91	1.82	SEM[2]	L	Q
Ruminal pH	6.24	6.20	6.42	0.06	0.06	0.12
Ruminal NH_3,[4] m*M*	1.93	3.85	5.43	0.31	<0.001	0.66
Fluid passage rate, %/h	5.74	6.86	6.47	0.45	0.28	0.19
Ruminal total VFA, m*M*	80.0	82.3	75.3	2.0	0.12	0.07
Ruminal VFA, mol/100 mol						
Acetate	74.5	73.1	68.6	0.3	<0.001	<0.001
Propionate	15.6	16.9	20.3	0.2	<0.001	<0.001
Butyrate	8.07	7.79	8.17	0.11	0.55	0.03
Isobutyrate	0.646	0.765	0.943	0.025	<0.001	0.35
Isovalerate	0.62	0.80	1.20	0.04	<0.001	0.03
Valerate	0.505	0.583	0.703	0.011	<0.001	0.12
Acetate:propionate	4.81	4.34	3.42	0.11	<0.001	0.10

[1]Treatments: 0 = ad libitum chopped native grass hay only; 0.91 = ad libitum chopped native grass hay plus 0.91 kg/d whole flaxseed (DM basis); 1.82 = ad libitum chopped native grass hay plus 1.82 kg/d of whole flaxseed (DM basis).
[2]n = 9.
[3]L = linear; Q = quadratic.
[4]Treatment × time interaction = $P < 0.001$.

the 0.91 and 1.82 kg/d inclusion of flaxseed at 15 h postdosing but similar treatment differences at other time points. Overall, the main effects of dietary treatment showed that increasing levels of whole flaxseed increased ($P < 0.001$) ruminal NH_3 concentrations. According to in situ analysis, 67.3% of flaxseed CP is effectively degradable (Mustafa et al., 2003). Ruminal NH_3 levels reported herein were above the minimum level (1.4 m*M*) suggested by Satter and Slyter (1974) to support microbial growth. No differences ($P = 0.19$) were observed for fluid passage rate across dietary treatments. This appears logical due to the lack of differences in total intake and ruminal OM digestibility, both of which can affect ruminal passage rate (Galyean and Owens, 1991). A quadratic tendency ($P = 0.07$) was observed for total ruminal VFA concentrations with cattle receiving 0.91 kg/d having the greatest concentrations and 1.82 kg/d of flaxseed being lowest in VFA concentration. Of the VFA measured, only isovalerate exhibited a treatment × time interaction ($P = 0.02$) at 3 h postdosing, which was due to a reduction in magnitude of the difference between 0.91 kg/d and control treatments (data not shown). The molar proportions of acetate decreased (quadratic, $P < 0.001$), whereas propionate increased (quadratic, $P < 0.001$) with increasing levels of dietary whole flaxseed. Ruminal molar proportions of isobutyrate, isovalerate, and valerate increased linearly ($P < 0.001$) as intake of whole flaxseed increased from 0 to 1.82 kg/d. The tendency of ruminal pH to increase with the addition of flaxseed is due to the trend for lower ruminal total VFA concentration as cattle consumed more flaxseed. Reduced total VFA production has been observed previously by Murphy et al. (1987) when increasing levels of rapeseed were fed to dairy cows. As with our results, Ikwuegbu and Sutton (1982) also observed an increase in propionate with increasing levels of linseed oil. The increased molar proportion of branched-chain VFA (isobutyrate, isovalerate) observed in this experiment with whole flaxseed supplementation is due to the increased ruminal supply of branched-chain AA (valine and leucine; Maeng and Baldwin, 1976). Mustafa et al. (2003) reported that leucine and valine in ground flaxseed had a ruminal digestibility of 83.5 and 79.9%, respectively, which would increase ruminal branched-chain VFA concentrations.

Fatty Acid Intake and Disappearance

Due to experimental design, fatty acid intake increased linearly ($P < 0.001$) for all dietary fatty acids measured (Table 6). Overall, total unsaturated fatty acid intake (18:1n-9, 18:2n-6, and 18:3n-3) increased from 33.4 to 442.1 g/d as whole flaxseed supplementation increased from 0 to 1.82 kg/d.

Ruminal biohydrogenation (percentage of 18C intake) of 18:1n-9 increased quadratically ($P = 0.004$) across treatments yet did not differ ($P \geq 0.33$) for 18:2n-6 and 18:3n-3 (Table 7). Furthermore, total 18C fatty acid biohydrogenation was not different ($P = 0.51$) when cattle were fed increasing amounts of whole flaxseed. Wu and Palmquist (1991) observed an increased in vitro biohydrogenation of 18:1n-9 when diets were supplemented with 3% Ca-soaps and biohydrogenation decreased when dietary Ca-soaps increased to 6%. Biohydrogenation intermediates expressed as either the proportion of dietary or duodenal 18C fatty acids responded quadratically ($P = 0.01$ and 0.001, respectively) with cattle consuming 0.91 kg/d of whole flaxseed having the greatest proportion of biohydrogenation intermediates compared with 0 or 1.82 kg/d. This response was unexpected given the fact that Harfoot et al. (1973) observed an irreversible inhibition of biohydrogenation and subsequent accumulation of biohydrogenation intermediates with high levels of linoleic

Table 6. Influence of supplemental whole flaxseed level (kg/d) on fatty acid intake (g/d) in beef heifers consuming native grass hay

	Treatment[1]				Contrast[3]	
Fatty acid	0	0.91	1.82	SEM[2]	L	Q
16:0	21.1	32.2	43.1	1.5	<0.001	0.96
18:0	2.6	10.1	17.6	0.4	<0.001	0.98
18:1n-9	5.0	43.3	81.6	0.8	<0.001	0.99
18:2n-6	11.9	50.0	88.4	1.4	<0.001	0.96
18:3n-3	16.4	144.2	272.1	1.9	<0.001	0.99
Total SFA[4]	23.8	42.3	60.7	1.8	<0.001	0.97
MUFA[5]	5.0	43.3	81.6	0.8	<0.001	0.99
PUFA[6]	28.3	194.3	360.5	3.3	<0.001	0.98
Total unsaturated fatty acids[7]	33.4	237.6	442.1	4.1	<0.001	0.98
Total	60.5	284.6	508.7	5.8	<0.001	1.00

[1]Treatments: 0 = ad libitum chopped native grass hay only; 0.91 = ad libitum chopped native grass hay plus 0.91 kg/d whole flaxseed (DM basis); 1.82 = ad libitum chopped native grass hay plus 1.82 kg/d of whole flaxseed (DM basis).
[2]n = 9.
[3]L = linear; Q = quadratic.
[4]Total SFA = 16:0 + 18:0.
[5]MUFA = 18:1n-9.
[6]PUFA = 18:2n-6 + 18:3n-3.
[7]Total unsaturated fatty acids = MUFA + PUFA.

acid. Likewise, Beam et al. (2000) found that for every percentage unit increase in linoleic acid there was a 1.2%/h decline in the rate of biohydrogenation. The reduction in biohydrogenation intermediates was due to the observed reduction in biohydrogenation of 18:1n-9, which led to a reduction in the accumulation of biohydrogenation intermediates when cattle were fed 1.82 kg/d of whole flaxseed. Although this experiment was not focused on examining the effects of feeding whole seeds as it relates to processing per se, we had hoped to provide some protection from biohydrogenation by feeding the whole seed, which did not appear to be the case as evidenced by the fairly extensive biohydrogenation of dietary 18C fatty acids. This agrees with Keele et al. (1989) who concluded that feeding whole cottonseed did not provide unsaturated fatty acids with much protection from biohydrogenation.

Duodenal supply of 18C fatty acids increased ($P < 0.001$) with increasing levels of whole flaxseed (Table 8). Duodenal flow of 17:0, 18:0, and 24:0 increased quadratically ($P \leq 0.04$), whereas 12:0, 16:0, and 20:0 increased linearly ($P \leq 0.05$) with increasing supply of whole flaxseed. The approximate 20-fold increase in intestinal supply of 18:0 compared with intake was due to biohydrogenation of 18C fatty acids averaging 82.1% across treatments. Significant increases in SFA may be concerning at first glance, especially because diets high in saturated fats are considered a detriment to human health. However, 18:0 has been shown to be as beneficial as 18:1n-9 in reducing plasma choles-

Table 7. Influence of supplemental whole flaxseed level (kg/d) on ruminal biohydrogenation[1] in beef heifers consuming native grass hay

	Treatment[2]				Contrast[4]	
Item	0	0.91	1.82	SEM[3]	L	Q
18:1n-9, % of 18:1n-9 intake	32.7	54.5	50.0	3.1	0.001	0.004
18:2n-6, % of 18:2n-6 intake	80.6	81.5	80.8	1.9	0.95	0.72
18:3n-3, % of 18:3n-3 intake	87.2	86.3	84.7	1.9	0.33	0.90
Total, % of C18 intake[5]	81.7	83.1	81.5	1.8	0.92	0.51
Biohydrogenation intermediates, proportion of duodenal C18 fatty acids	4.80	8.88	7.58	0.56	0.002	0.001
Biohydrogenation intermediates, proportion of dietary C18 fatty acids	2.22	3.96	3.39	0.24	<0.001	0.01

[1]Biohydrogenation was calculated using the equations reported by Scholljegerdes et al. (2004a).
[2]Treatments: 0 = ad libitum chopped native grass hay only; 0.91 = ad libitum chopped native grass hay plus 0.91 kg/d whole flaxseed (DM basis); 1.82 = ad libitum chopped native grass hay plus 1.82 kg/d of whole flaxseed (DM basis).
[3]n = 9.
[4]L = linear; Q = quadratic.
[5]Total = 18:1n-9 + 18:2n-6 + 18:3n-3.

Table 8. Influence of supplemental whole flaxseed level (kg/d) on duodenal fatty acid flow (g/d) in beef heifers consuming native grass hay

	Treatment[1]				Contrast[3]	
Fatty acid	0	0.91	1.82	SEM[2]	L	Q
12:0	0.74	0.80	0.89	0.05	0.05	0.83
14:0	2.77	4.33	3.94	1.07	0.45	0.47
14:1	2.64	2.62	2.39	0.08	0.04	0.26
15:0	2.53	2.68	2.69	0.37	0.75	0.84
15:1	1.83	0.07	0.16	0.94	0.23	0.43
16:0	21.3	36.1	84.5	8.4	<0.001	0.12
16:1	1.03	0.89	0.78	0.08	0.05	0.87
17:0	1.38	1.78	1.64	0.08	0.04	0.02
18:0	43	251	366	19	<0.001	<0.001
18:1 *trans*-9	0.00	2.46	2.36	0.16	<0.001	<0.001
18:1 *trans*-11	2.7	13.9	15.5	1.3	<0.001	0.01
18:1 *trans*-13+14	0.0	13.7	17.5	1.3	<0.001	0.004
18:1n-9	5.4	27.3	46.2	2.9	<0.001	0.66
18:2 *trans*-9 *trans*-12	0.00	1.14	1.67	0.20	<0.001	0.20
18:2n-6	3.5	12.7	19.9	2.1	<0.001	0.67
20:0	2.16	2.27	2.74	0.19	0.05	0.46
18:3n-3	3.2	27.2	50.4	6.3	<0.001	0.95
20:1	0.39	0.67	0.27	0.13	0.52	0.04
22:0	1.91	2.22	1.80	0.29	0.76	0.25
20:3n-6	0.00	0.00	0.11	0.06	0.24	0.49
23:0	0.27	0.35	0.32	0.06	0.59	0.51
22:2	0.59	0.31	0.61	0.22	0.92	0.23
24:0	1.89	2.42	1.58	0.31	0.40	0.04
20:5n-3	0.58	0.65	0.13	0.32	0.05	0.34
24:1	0.26	0.02	0.30	0.11	0.77	0.07
Other	17.9	39.6	43.3	2.9	<0.001	0.02
Total SFA[4]	78	305	466	21	<0.001	0.23
MUFA[5]	13.9	61.0	85.3	3.7	<0.001	0.02
PUFA[6]	8.2	42.2	74.7	8.3	<0.001	0.93
Total unsaturated fatty acids[7]	22.1	103.2	160.0	10.7	<0.001	0.29
Total	117	447	669	28	<0.001	0.14

[1]Treatments: 0 = ad libitum chopped native grass hay only; 0.91 = ad libitum chopped native grass hay plus 0.91 kg/d whole flaxseed (DM basis); 1.82 = ad libitum chopped native grass hay plus 1.82 kg/d of whole flaxseed (DM basis).
[2]n = 9.
[3]L = linear; Q = quadratic.
[4]Total SFA = 12:0 + 14:0 + 15: 0 + 16:0 + 17:0 + 18:0 + 20:0 + 22:0 + 23:0 + 24:0.
[5]MUFA = 14:1 + 15:1 + 16:1 + 18:1 *trans*-9 + 18:1 *trans*-11 + 18:1 *trans*-13+14 + 18:1n-9 + 20:1 + 24:1.
[6]PUFA = 18:2 *trans*-9 *trans*-12 + 18:2n-6 + 18:3n-3 + 20:3n-6 + 22:2 + 20:5n-3.
[7]Total unsaturated fatty acids = MUFA + PUFA.

terol levels compared with other SFA (Bonanome and Grundy, 1988). Duodenal supply of all 18:1 isomers increased ($P < 0.001$) in flax-fed cattle compared with unsupplemented controls and there was a quadratic increase ($P < 0.01$) for 18:1 *trans*-9, 18:1 *trans*-11, and 18:1 *trans*-13+14, whereas 18:1n-9 increased linearly ($P < 0.001$) with increasing levels of whole flaxseed. This agrees with Loor et al. (2004) who reported an increase in duodenal supply of 18:1 isomers when linseed oil was added to either a low concentrate or high concentrate dairy-type diet. Flax feeding increased ($P < 0.001$) the intestinal supply of 18:3n-3. Intestinal supply of MUFA responded quadratically ($P = 0.02$) to increasing levels of flaxseed. Intestinal PUFA and total unsaturated fatty acid supply increased linearly ($P < 0.001$) with flaxseed supplementation. These data agree with those of others (Murphy et al., 1987; Wu et al., 1991; Kucuk et al., 2004) in which intestinal supply of MUFA and PUFA increased with a greater fatty acid intake.

Although our primary objective was to evaluate intestinal supply of fatty acids, it is important to know the extent to which those fatty acids are available postruminally. Apparent postruminal disappearance of 14:1, 18:1 *trans*-13+14, and 18:3n-3 tended ($P = 0.06$ to 0.08) to decline, whereas 12:0 increased in digestibility with level of whole flaxseed (Table 9). Apparent postruminal disappearance of 18:0, 18:1n-9, 18:2n-6, total SFA, and MUFA decreased linearly ($P = 0.001$ to 0.04) across dietary treatments. The decrease in intestinal digestibility with increasing levels of dietary fat has been reported previously by others (Doreau and Ferlay, 1994; Plascencia et al., 2003; Kucuk et al., 2004). Intestinal digestibility is generally greater for unsaturated fatty acids compared with SFA due to their ability to easily form micelles in the small intestine (Ockner et al.,

Table 9. Influence of supplemental whole flaxseed level (kg/d) on apparent postruminal fatty acid disappearance (% of duodenal flow) in beef heifers consuming native grass hay

	Treatment[1]				Contrast[3]	
Fatty acid	0	0.91	1.82	SEM[2]	L	Q
12:0	73.3	82.3	83.5	3.6	0.06	0.31
14:0	64.5	66.6	72.5	4.1	0.15	0.66
14:1	81.9	74.2	72.1	3.7	0.06	0.49
15:0	70.1	71.0	70.6	3.8	0.93	0.88
16:0	72.4	63.4	71.2	4.6	0.84	0.11
16:1	68.1	59.9	62.3	7.8	0.77	0.40
17:0	68.2	60.7	57.3	6.3	0.21	0.78
18:0	86.6	69.6	65.3	3.7	0.001	0.16
18:1 *trans*-11	88.7	86.2	85.5	2.7	0.37	0.76
18:1 *trans*-13+14	10.4	87.2	87.2	2.3	0.06	0.08
18:1n-9	73.2	45.7	46.9	6.0	0.01	0.06
18:2n-6	73.4	52.7	48.1	7.2	0.02	0.34
20:0	61	24	45	14	0.42	0.12
18:3n-3	65.7	55.4	48.6	6.9	0.07	0.82
24:0	34.6	30.5	0.0	20	0.24	0.57
Total SFA[4]	78.1	68.1	66.5	3.6	0.04	0.34
MUFA[5]	80.1	66.0	64.9	4.2	0.03	0.20
PUFA[6]	56.3	51.5	50.9	5.9	0.45	0.73
Total unsaturated fatty acids[7]	72.0	63.3	63.1	4.2	0.14	0.40
Total	74.3	67.7	66.9	3.2	0.12	0.46

[1]Treatments: 0 = ad libitum chopped native grass hay only; 0.91 = ad libitum chopped native grass hay plus 0.91 kg/d whole flaxseed (DM basis); 1.82 = ad libitum chopped native grass hay plus 1.82 kg/d of whole flaxseed (DM basis).
[2]n = 9.
[3]L = linear; Q = quadratic.
[4]Total SFA = 12:0 + 14:0 + 15: 0 + 16:0 + 17:0 + 18:0 + 20:0 + 22:0 + 23:0 + 24:0.
[5]MUFA = 14:1 + 15:1 + 16:1 + 18:1 *trans*-9 + 18:1 *trans*-11 + 18:1 *trans*-13+14 + 18:1n-9 + 20:1 + 24:1.
[6]PUFA = 18:2 *trans*-9 *trans*-12 + 18:2n-6 + 18:3n-3 + 20:3n-6 + 22:2 + 20:5n-3.
[7]Total unsaturated fatty acids = MUFA + PUFA.

1972). Values for intestinal digestibility of 18:0 were within range of others (Aldrich et al., 1997; Wachira et al., 2000; Scholljegerdes et al., 2004a) who have fed oilseeds. According to the extensive review by Doreau and Chilliard (1997a), intestinal digestibility of fatty acids ranged from 55 to 92%. These differences among fatty acids are generally due to chain length and number of double bonds (Doreau and Chilliard, 1997a). In the current study, apparent intestinal digestibility of 18:3n-3 ranged from 65.7 to 48.6% as intake of flaxseed increased from 0 to 1.82 kg/d. Wachira et al. (2000) reported much greater intestinal digestibility of 18:3n-3 (91.0% of duodenal flow) in sheep fed a 71.5% forage diet supplemented with whole flaxseed. Those diets were formulated to provide 4.8% total fatty acids, which is similar to that of cattle fed 0.91 kg/d whole flaxseed (4.1% total fatty acids) in the current experiment, ruling out dietary fatty acid concentration as a reason. The large discrepancy between Wachira et al. (2000) and the current experiment may be the inherent differences in extent of mastication between sheep and cattle. As with our data, Aldrich et al. (1997) noted relatively low values for postruminal supply of 18C fatty acids when steers were fed whole canola seed. These relatively low values for postruminal digestibility could be due to some protection of fatty acids by the seed coat, which caused fatty acids to be less available in the small intestine. In addition, hind gut fermentation and microbial fatty acid production may have elevated fatty acid concentration in feces, thus lowering postruminal fatty acid digestibility.

Several reviews have outlined positive effects of supplemental fatty acids on the composition of ruminant products (Palmquist and Jenkins, 1980; Wood and Enser, 1997; Demeyer and Doreau, 1999) or as precursors for reproductive hormones (Staples et al., 1998; Abayasekara and Wathes, 1999; Funston, 2004). However, these advantages often come at the expense of intake, diet digestibility, or growth performance. Therefore, it is important to consider trade-offs of fat-supplemented diets between known physiological functions and animal performance. In the current trial, flaxseed increased the supply of intestinal fatty acids and may increase tissue supply of unsaturated fatty acids when fed under the conditions described herein. However, more work is needed in this area to elucidate the efficacy of flaxseed for improving growth performance, tissue fatty acid composition, and reproductive success in forage-based production systems.

LITERATURE CITED

Abayasekara, D. R., and D. C. Wathes. 1999. Effects of altering dietary fatty acid composition on prostaglandin synthesis and fertility. Prostaglandins Leukot. Essent. Fatty Acids 61:275–287.

Albro, J. D., D. W. Weber, and T. DelCurto. 1993. Comparison of whole, raw soybeans, extruded soybeans, or soybean meal and barley on digestive characteristics and performance of weaned beef steers consuming mature grass hay. J. Anim. Sci. 71:23–32.

Aldrich, C. G., N. R. Merchen, J. K. Drackley, S. S. Gonzalez, G. C. Fahey Jr., and L. L. Berger. 1997. The effects of chemical treatment of whole canola seed on lipid and protein digestion by steers. J. Anim. Sci. 75:502–511.

Allen, M. S. 2000. Effects of diet on short-term regulation of feed intake by lactating dairy cattle. J. Dairy Sci. 83:1598–1624.

Ambrose, D. J., J. P. Kastelic, R. Corbett, P. A. Pitney, H. V. Petit, J. A. Small, and P. Zalkovic. 2006. Lower pregnancy losses in lactating dairy cows fed a diet enriched in alpha-linolenic acid. J. Dairy Sci. 89:3066–3074.

AOAC. 1990. Official Methods of Analysis. 15th ed. Assoc. Off. Anal. Chem., Arlington, VA.

Beam, T. M., T. C. Jenkins, P. J. Moate, R. A. Kohn, and D. L. Palmquist. 2000. Effects of amount and source of fat on the rates of lipolysis and biohydrogenation of fatty acids in ruminal contents. J. Dairy Sci. 83:2564–2573.

Bonanome, A., and S. M. Grundy. 1988. Effect of dietary stearic acid on plasma cholesterol and lipoprotein levels. N. Engl. J. Med. 318:1244–1248.

Broderick, G. A., and J. H. Kang. 1980. Automated simultaneous determination of ammonia and total amino acids in ruminal fluid and in vitro media. J. Dairy Sci. 63:64–75.

Brokaw, L., B. W. Hess, and D. C. Rule. 2001. Supplemental soybean oil or corn for beef heifers grazing summer pasture: Effects on forage intake, ruminal fermentation, and site and extent of digestion. J. Anim. Sci. 79:2704–2712.

Demeyer, D., and M. Doreau. 1999. Targets and procedures for altering ruminant meat and milk lipids. Proc. Nutr. Soc. 58:593–607.

Doreau, M., and Y. Chilliard. 1997a. Digestion and metabolism of dietary fat in farm animals. Br. J. Nutr. 78:S15–S35.

Doreau, M., and Y. Chilliard. 1997b. Effects of ruminal or postruminal fish oil supplementation on intake and digestion in dairy cows. Reprod. Nutr. Dev. 37:113–124.

Doreau, M., and A. Ferlay. 1994. Digestion and utilization of fatty acids by ruminants. Anim. Feed Sci. Technol. 45:379–396.

Drouillard, J. S., M. A. Seyfert, E. J. Good, E. R. Loe, B. Depenbusch, and R. Daubert. 2004. Flaxseed for finishing beef cattle: Effects on animal performance, carcass quality, and meat composition. Pages 108–117 in Proc. 60th Flax Inst., Fargo, ND. Flax Inst., Dept. Plant Sci., Fargo, ND.

Funston, R. N. 2004. Fat supplementation and reproduction in beef females. J. Anim. Sci. 82(E Suppl.):E154–E161.

Galyean, M. L., and F. N. Owens. 1991. Effects of diet composition and level of feed intake on site and extent of digestion in ruminants. Pages 483–513 in Physiological Aspects of Digestion and metabolism in Ruminants. T. Tsuda, Y. Sasaki, and R. Kawashima, ed. Academic Press, Toronto.

Goetsch, A. L., and M. L. Galyean. 1983. Influence of feeding frequency on passage of fluid and particulate markers in steers fed a concentrate diet. Can. J. Anim. Sci. 63:727–730.

Gonthier, C., A. F. Mustafa, R. Berthiaume, H. V. Petit, R. Martineau, and D. R. Ouellet. 2004. Effects of feeding micronized and extruded flaxseed on ruminal fermentation and nutrient utilization by dairy cows. J. Dairy Sci. 87:1854–1863.

Harfoot, C. G., R. C. Noble, and J. H. Moore. 1973. Factors influencing the extent of biohydrogenation of linoleic acid by rumen micro-organisms in vitro. J. Sci. Food Agric. 24:961–970.

Ikwuegbu, O. A., and J. D. Sutton. 1982. The effect of varying the amount of linseed oil supplementation on rumen metabolism in sheep. Br. J. Nutr. 48:365–375.

Jenkins, T. C. 1993. Lipid metabolism in the rumen. J. Dairy Sci. 76:3851–3863.

Keele, J. W., R. E. Roffler, and K. Z. Beyers. 1989. Ruminal metabolism in nonlactating cows fed whole cottonseed or extruded soybeans. J. Anim. Sci. 67:1612–1622.

Kronberg, S. L., G. Barceló-Coblijn, J. Shin, K. Lee, and E. J. Murphy. 2006. Bovine muscle n-3 fatty acid content is increased with flaxseed feeding. Lipids 41:1059–1068.

Kucuk, O., B. W. Hess, P. A. Ludden, and D. C. Rule. 2001. Effect of forage:concentrate ratio on ruminal digestion and duodenal flow of fatty acids in ewes. J. Anim. Sci. 79:2233–2240.

Kucuk, O., B. W. Hess, and D. C. Rule. 2004. Soybean oil supplementation of a high-concentrate diet does not affect site and extent of organic matter, starch, neutral detergent fiber, or nitrogen digestion, but influences both ruminal metabolism and intestinal flow of fatty acids in limit-fed lambs. J. Anim. Sci. 82:2985–2994.

Lake, S. L., E. J. Scholljegerdes, T. R. Weston, D. C. Rule, and B. W. Hess. 2006. Postpartum supplemental fat, but not maternal body condition score at parturition, affects plasma and adipose tissue fatty acid profiles of suckling beef calves. J. Anim. Sci. 84:1811–1819.

Leupp, J. L., G. P. Lardy, S. A. Soto-Navarro, M. L. Bauer, and J. S. Caton. 2006. Effects of canola seed supplementation on intake, digestion, duodenal protein supply, and microbial efficiency in steers fed forage-based diets. J. Anim. Sci. 84:499–507.

Loor, J. J., K. Ueda, A. Ferlay, Y. Chilliard, and M. Doreau. 2004. Biohydrogenation, duodenal flow, and intestinal digestibility of *trans* fatty acids and conjugated linoleic acids in response to dietary forage:concentrate ratio and linseed oil in dairy cows. J. Dairy Sci. 87:2472–2485.

Maddock, T. D., M. L. Bauer, K. B. Koch, V. L. Anderson, R. J. Maddock, G. Barceló-Coblijn, E. J. Murphy, and G. P. Lardy. 2006. Effect of processing flax in beef feedlot diets on performance, carcass characteristics, and trained sensory panel ratings. J. Anim. Sci. 84:1544–1551.

Maeng, W. J., and R. L. Baldwin. 1976. Factors influencing rumen microbial growth rates and yields: Effects of amino acid addition to a purified diet with N from urea. J. Dairy Sci. 59:648–655.

Merchen, N. R., J. L. Firkins, and L. L. Berger. 1986. Effect of intake and forage level on ruminal turnover rates, bacterial protein synthesis and duodenal amino acid flow in sheep. J. Anim. Sci. 62:216–225.

Moore, J. A., R. S. Swingle, and W. H. Hale. 1986. Effects of whole cottonseed, cottonseed oil or animal fat on digestibility of wheat straw diets by steers. J. Anim. Sci. 63:1267–1273.

Murphy, M., P. Udén, D. L. Palmquist, and H. Wiktorsson. 1987. Rumen and total diet digestibilities in lactating cows fed diets containing full-fat rapeseed. J. Dairy Sci. 70:1572–1582.

Mustafa, A. F., C. Gonthier, and D. R. Ouellet. 2003. Effects of extrusion of flaxseed on ruminal and postruminal nutrient digestibilities. Arch. Anim. Nutr. 57:455–463.

Myers, W. D., P. A. Ludden, V. Nayigihugu, and B. W. Hess. 2004. Technical Note: A procedure for the preparation and quantitative analysis of samples for titanium dioxide. J. Anim. Sci. 82:179–183.

Ockner, R. K., J. P. Pitman, and J. L. Yager. 1972. Differences in the intestinal absorption of saturated and unsaturated long chain fatty acids. Gastroenterology 62:981–992.

Palmquist, D. L., and T. C. Jenkins. 1980. Review: Fat in lactation rations. J. Dairy Sci. 63:1–14.

Pavan, E., S. K. Duckett, and J. G. Andrae. 2007. Corn oil supplementation to steers grazing endophyte-free tall fescue. I. Effects on in vivo digestibility, performance, and carcass traits. J. Anim. Sci. 85:1330–1339.

Plascencia, A., G. D. Mendoza, C. Vásquez, and R. A. Zinn. 2003. Relationship between body weight and level of fat supplementation on fatty acid digestion in feedlot cattle. J. Anim. Sci. 81:2653–2659.

Satter, L. D., and L. L. Slyter. 1974. Effect of ammonia concentration of rumen microbial protein production in vitro. Br. J. Nutr. 32:199–208.

Schauff, D. J., and J. H. Clark. 1992. Effects of feeding diets containing calcium salts of long-chain fatty acids to lactating dairy cows. J. Dairy Sci. 75:2990–3002.

Scholljegerdes, E. J., B. W. Hess, G. E. Moss, D. L. Hixon, and D. C. Rule. 2004a. Influence of supplemental cracked high-linoleate or high-oleate safflower seeds on site and extent of digestion in beef cattle. J. Anim. Sci. 82:3577–3588.

Scholljegerdes, E. J., P. A. Ludden, and B. W. Hess. 2004b. Site and extent of digestion and amino acid flow to the small intestine in beef cattle consuming limiting amounts of forage. J. Anim. Sci. 82:1146–1156.

Soita, H. W., J. A. Meier, M. Fehr, P. Yu, D. A. Christensen, J. J. McKinon, and A. F. Mustafa. 2003. Effects of flaxseed supplementation on milk production, milk fatty acid composition and nutrient utilization by lactating dairy cows. Arch. Anim. Nutr. 57:107–116.

Staples, C. R., J. M. Burke, and W. W. Thatcher. 1998. Influence of supplemental fats on reproductive tissues and performance of lactating cows. J. Dairy Sci. 81:856–871.

Steel, R. G. D., and J. H. Torrie. 1980. Principles and Procedures of Statistics: A Biometrical Approach. 2nd ed. McGraw-Hill Book Co., New York, NY.

Tice, E. M., M. L. Eastridge, and J. L. Firkins. 1994. Raw soybeans and roasted soybeans of different particle sizes. 2. Fatty acid utilization by lactating dairy cows. J. Dairy Sci. 77:166–180.

Uden, P., P. E. Colucci, and P. J. Van Soest. 1980. Investigations of chromium, cerium, cobalt as markers in digesta. Rate of passage studies. J. Sci. Food Agric. 31:625–632.

Ueda, K., A. Ferlay, J. Chabrot, J. J. Loor, Y. Chilliard, and M. Doreau. 2003. Effect of linseed oil supplementation on ruminal digestion in dairy cows fed diets with different forage:concentrate ratios. J. Dairy Sci. 86:3999–4007.

Wachira, A. M., L. A. Sinclair, R. G. Wilkinson, K. Hallett, M. Enser, and J. D. Wood. 2000. Rumen biohydrogenation of n-3 polyunsaturated fatty acids and their effects on microbial efficiency and nutrient digestibility in sheep. J. Agric. Sci. 135:419–428.

Weill, P., B. Schmitt, G. Chesneau, N. Daniel, F. Safraou, and P. Legrand. 2002. Effects of introducing linseed in livestock diet on blood fatty acid composition of consumers of animal products. Ann. Nutr. Metab. 46:182–191.

Whitney, M. B., B. W. Hess, J. E. Kaltenbach, H. J. Harlow, and D. C. Rule. 1999. Direct transesterification of lipids from feedstuffs and ruminal bacteria. Can. J. Anim. Sci. 79:247–249.

Wood, J. D., and M. Enser. 1997. Factors influencing fatty acids in meat and the role of antioxidants in improving meat quality. Br. J. Nutr. 78:S49–S60.

Wu, Z., O. A. Ohajuruka, and D. L. Palmquist. 1991. Ruminal synthesis, biohydrogenation, and digestibility of fatty acids in dairy cows. J. Dairy Sci. 74:3025–3034.

Wu, Z., and D. L. Palmquist. 1991. Synthesis and biohydrogenation of fatty acid by ruminal microorganisms in vitro. J. Dairy Sci. 74:3035–3046.

Zhang, R. H., A. F. Mustafa, and X. Zhao. 2007. Effects of feeding oilseeds on nutrient utilization by lactating ewes. Small Rumin. Res. 67:307–311.

Zinn, R. A., and F. N. Owens. 1986. A rapid procedure for purine measurement and its use for estimating net ruminal protein synthesis. Can. J. Anim. Sci. 66:157–166.